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Burroughs Wellcome Co.

3030 Cornwallis Road
Research Triangle Park, N.C. 27709

Wellcome

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TWX5109270915
tel. 919 248-3000

September 28, 1990

Commissioner of Patents and Trademarks
Box Patent Ext.
Washington, DC 20231

Sir:

Please address all communications relating to the enclosed APPLICATION FOR EXTENSION OF PATENT TERM UNDER 35 U.S.C. 156 of U.S. Patent No. 4,312,860 to Dr. L. A. Nielsen, Burroughs Wellcome Co., 3030 Cornwallis Road, Research Triangle Park, NC 27709; telephone no. (919) 248-4126.

Very truly yours,

David A. Yeowell

David A. Yeowell, Ph.D.
Vice President - Technical Development

DAY:sls

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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Patent No.: 4,312,860
Issue Date: January 26, 1982
For: LUNG SURFACTANT COMPOSITIONS
Inventor: John A. Clements
Assignee: Regents of the University of California
Alameda, California

DECLARATION

To the Commissioner of Patents and Trademarks:

Burroughs Wellcome Co., a corporation organized under the laws of the State of North Carolina, having a place of business at 3030 Cornwallis Road, Research Triangle Park, North Carolina 27709 (hereinafter referred to as "Wellcome"), declares as follows:

1) That Wellcome makes this declaration as the special agent of The Regents of the University of California, organized under the laws of the State of California and having a business location at 1320 Harbor Bay Parkway, Suite 150, Alameda, California 94501 (herein after referred to as the "Regents").

2) That Wellcome has been authorized to act as the Regents' special agent for the sole and limited purpose of preparing, filing and prosecuting an application for the extension of the term of United States Patent 4,312,860, issued January 26, 1982 (hereinafter referred to as the "Patent") and to do every act in connection with the above stated purpose which Wellcome deems necessary or desirable.

3) That Wellcome believes the Regents are the assignee of the entire right, title and interest in the Patent.

4) That submitted herewith is an APPLICATION FOR EXTENSION OF PATENT TERM UNDER 35 U.S.C. 156 of the Patent (hereinafter referred to as the "Application") on behalf of the Regents requesting a 1029 day extension of the term of the Patent.

5) That Wellcome has reviewed and understands the contents of the Application which is submitted pursuant to 35 U.S.C. 156.

6) That Wellcome believes that the Patent is subject to extension pursuant to 37 CFR 1.710.

7) That Wellcome believes that a 1029 day extension of the term of the Patent is fully justified under 35 U.S.C. 156 and applicable regulations.

8) That Wellcome believes the Patent meets the conditions for the extension of the term of a patent as set forth 37 CFR 1.720.

Wellcome declares further that all statements made herein of its own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements are made with the knowledge that willful false statements and the like are punishable by fine or imprisonment, or both, under Section 1001 of title 18 of the United States Code and that such willful false statements may jeopardize the validity of United States Patent 4,312,860, issued January 26, 1982, and any extensions thereof.

Burroughs Wellcome Co., as Special Agent
for The Regents of the
University of California,
Alameda, California

By David A. Yeowell Date: 9/27/90

David A. Yeowell
Vice President, Technical Development
Burroughs Wellcome Co.

Docket No. 90/PD/407

"Express Mail" Label No. AB192629206

Date of Deposit Sept. 28, 1990

I hereby certify that this paper or fee is being deposited with the United States Postal Service "Express Mail Post Office to Addressee" service under 37 CRF 1.10 on the date indicated above and is addressed to the Commissioner of Patents and Trademarks, Box Patent Ext. Washington, DC 20231.

Lawrence A. Hur
(Reg. No. 29682)

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In Re: U.S. Patent No. 4,312,860

Issued: January 26, 1982

To: John A. Clements

For: LUNG SURFACTANT COMPOSITIONS

Commissioner of Patents and Trademarks

Box Patent Ext.

Washington, D.C. 20231

APPLICATION FOR EXTENSION OF PATENT TERM UNDER 35 U.S.C. 156

Sir:

THE REGENTS of THE UNIVERSITY OF CALIFORNIA, organized under the Laws of the State of California and having a business location at 1320 Harbor Bay Parkway, Suite 150, Alameda, California 94501 (hereinafter "Applicant"), represents that it is the assignee of the entire interest in and to

Letters Patent of the United States of America No. 4,312, 860 granted to John A. Clements, January 26, 1982 for LUNG SURFACTANT COMPOSITIONS, by virtue of an assignment to Applicant recorded in the United States Patent and Trademark Office on November 10, 1980, Reel 3805, Frames 877-879.

BURROUGHS WELLCOME CO., a corporation organized under the laws of the State of North Carolina (hereinafter "Wellcome"), having been authorized by Applicant by virtue of a Special Power of Attorney dated September 21, 1990, a duplicate original of which is attached hereto as EXHIBIT 1, to act as its special agent to apply for an extension of the term of United States Patent 4,312,860, hereby submits on behalf of Applicant this application for extension of patent term under 35 U.S.C. 156 by providing the following information pursuant to 37 CFR 1.740.. For convenience, the information contained in this application will be presented according to the format set forth in 37 CFR 1.740.

(1) This application for extension is based upon the regulatory review period before the Food and Drug Administration ("FDA") of Wellcome's approved product, EXOSURF® Neonatal™ (colfosceril palmitate, cetyl alcohol, tyloxapol) for Intratracheal Suspension (hereinafter "EXOSURF®"). The active ingredient in EXOSURF® is colfosceril palmitate.* The package insert approved by FDA as part of NDA 20-044 (described below) for the approved product is attached hereto. Applicant has exclusively licensed Wellcome to make, have made, use and sell EXOSURF® under United States Patent 4,312,860.

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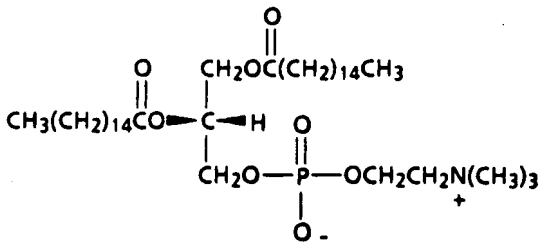
*The New Drug Application for EXOSURF® which was submitted by Wellcome identifies colfosceril palmitate as the only active ingredient. The FDA reviewed this submission pursuant to its single active ingredient policy. Due primarily to clinical concerns, the FDA clinical reviewers insisted that the ingredients cetyl alcohol and tyloxapol be prominently identified on the labeling. However, Wellcome continues to regard colfosceril palmitate as the sole active ingredient of EXOSURF®.

Colfosceril palmitate is designated chemically as dipalmitoylphosphatidylcholine and has the following chemical structure:



(2) The approved product was subject to regulatory review under Federal Food, Drug and Cosmetic Act, Section 505 (21 U.S.C. 355).

(3) EXOSURF® received permission for commercial marketing or use under Section 505 of the Federal Food, Drug and Cosmetic Act (21 U.S.C. 355) on August 2, 1990.

(4) Colfosceril palmitate is the active ingredient of EXOSURF®. Colfosceril palmitate has not been previously approved for commercial marketing under the Federal Food Drug and Cosmetic Act.

(5) This application for extension of patent term under 35 U.S.C. 156 is being submitted within the permitted 60 day period, which period will expire on October 1, 1990.

(6) The complete identification of the patent for which extension of term is being sought is as follows:

Inventors: John A. Clements

Patent Number: 4,312,860

Issue Date: January 26, 1982

(7) A complete copy of the patent identified in paragraph (6) above is appended hereto as EXHIBIT 2.

(8) There are no disclaimers, certificates of correction, maintenance fee payment receipts or reexamination certificates which relate to United States Patent No. 4,312,860.

(9) United States Patent Number 4,312,860 claims the approved product and a method of using the approved product. The patent claims applicable to the approved product are as follows:

Claim 1 reads as follows:

1. "A mammalian lung surfactant composition consisting essentially of dipalmitoyl phosphatidylcholine in admixture with a fatty alcohol."

The approved product consists essentially of dipalmitoylphosphatidylcholine in admixture with cetyl alcohol as well as a smaller amount of tyloxapol and sufficient sodium chloride to provide a suspension in saline solution upon reconstitution with Sterile Water for Injection.

Claim 2 reads as follows:

2. "The composition of claim 1 wherein the fatty alcohol has from about 14 to 18 carbon atoms."

The approved product consists essentially of dipalmitoylphosphatidylcholine in admixture with cetyl alcohol, a fatty alcohol which contains 16 carbon atoms, as well as a smaller amount of tyloxapol and sufficient sodium chloride to provide a suspension in saline solution upon reconstitution with Sterile Water for Injection.

Claim 3 reads as follows:

3. "The composition of claim 2 wherein the fatty alcohol is hexadecanol."

The approved product consists essentially of dipalmitoylphosphatidylcholine in admixture with 1-hexadecanol, which is an alternate name for cetyl alcohol, as well as a smaller amount of tyloxapol and sufficient sodium chloride to provide a suspension in saline solution upon reconstitution with Sterile Water for Injection.

Claim 5 reads as follows:

5. "The composition of claim 1 wherein the dipalmitoyl phosphatidyl choline constitutes a major percentage by weight of the composition and wherein the fatty alcohol constitutes a minor percentage."

The approved product contains dipalmitoylphosphatidylcholine (108 mg per 10 mL vial) and cetyl alcohol (12 mg per 10 mL vial).

Claim 6 reads as follows:

6. "The composition of claim 5 wherein the fatty alcohol is present in the range of about 6 to 18% by weight and the dipalmitoyl phosphatidyl choline is present in the range of about 82 to 94% by weight."

In the approved product, dipalmitoylphosphatidylcholine comprises approximately 84.4% of the total organic chemical components and cetyl alcohol comprises approximately 9.4% of the total organic chemical components.

Claim 7 reads as follows:

7. "A composition for administration into mammalian alveolar spaces comprising a suspension of dipalmitoyl phosphatidyl choline and hexadecanol in saline solution."

When reconstituted with Sterile Water for Injection, the approved product provides a suspension of dipalmitoylphophatidylcholine and cetyl alcohol (hexadecanol) as well as a smaller amount of tyloxapol in saline solution.

Claim 8 reads as follows:

8. "A method for treating respiratory distress syndrome in mammals wherein natural lung surfactant normally produced by the mammal is absent or deficient, comprising introducing into the alveolar spaces a quantity of a composition consisting essentially of a major amount of 1,2 dipalmitoyl-sn-3-glycerophosphoryl choline in admixture with a minor amount of a fatty alcohol."

The approved product, which comprises a composition consisting essentially of a major amount of 1,2- dipalmitoyl-sn-3-glycerophosphoryl choline (an alternate name for dipalmitoylphosphatidylcholine) in admixture with a minor amount of cetyl alcohol (a fatty alcohol) and a smaller amount of tyloxapol, is indicated for the treatment of infants who have developed or are at risk of developing respiratory distress syndrome or who have evidence of pulmonary insufficienty and is administered intratracheally as a reconstituted suspension in saline solution.

Claim 9 reads as follows:

"The method of claim **8** wherein the fatty alcohol is n-hexadecan-1-ol."

The approved product, which comprises a composition consisting essentially of a major amount of dipalmitoylphosphatidylcholine in admixture with a minor amount of n-hexadecan-1-ol (an alternate name for cetyl alcohol) and a smaller amount of tyloxapol, is indicated for the treatment of infants who have developed or are at risk of developing respiratory distress syndrome or who have evidence of pulmonary insufficienty and is administered intratracheally as a reconstituted aqueous suspension.

(10) The relevant dates and information pursuant to 35 U.S.C. 156(g) necessary to enable the Secretary of Health and Human Services to determine the applicable regulatory review period are as follows:

(a) U.S. Patent No. 4,312,860 was issued January 26, 1982.

(b) An investigational New Drug Application ("IND") for a lung surfactant composition (i.e., EXOSURF®) was filed by Roderick Phibbs, MD, Professor of Pediatrics and Chief of Neonatology at the University of California, San Francisco on May 1, 1985 as IND 26,298* and became effective May 31, 1985.

(c) An IND for EXOSURF® was filed by Wellcome on March 4, 1986 as IND 28,006 and became effective April 3, 1986.

(d) A New Drug Application ("NDA") for EXOSURF® was submitted by Wellcome on February 16, 1990 as NDA 20-044.

(e) NDA 20-044 for EXOSURF® was approved by the FDA on August 2, 1990.

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*Wellcome's Drug Regulatory Affairs Department assisted Dr. Phibbs in preparing IND 26,298. In addition, study results of clinical investigations conducted under IND 26,298 were included in IND 28,006 which was submitted by Wellcome. Therefore, Applicant considers that IND 26,298 should be considered for purposes of determining the regulatory review period.

(11) As a brief description of the activities undertaken by Phibbs and Wellcome during the applicable regulatory review period, attached hereto as EXHIBIT 3, is a chronology of the major communications between the Phibbs or Wellcome, as applicable, and the FDA from May 1, 1985 to August 2, 1990.

(12) Wellcome, as special agent for Applicant, is of the opinion that U.S. Patent 4,312,860 is eligible for extension under 35 U.S.C. 156 because it satisfies all the requirements for such extensions as follows:

(a) 35 U.S.C. 156(a)

U.S. Patent 4,312,860 claims a product and a method of using a product.

(b) 35 U.S.C. 156(a)(1)

The term of U.S. Patent 4,312,860 has not expired before submission of this application.

(c) 35 U.S.C. 156 (a)(2)

The term of U.S. Patent 4,312,860 has never been extended.

(d) 35 U.S.C. 156 (a)(3)

The application for extension is submitted by the owner of record through its agent in accordance with the requirements of 35 U.S.C. 156(d) and 37 CFR 1.710 et. seq.

(e) 35 U.S.C. 156 (a)(4)

The approved product has been subject to a regulatory review period before its commercial marketing or use.

(f) 35 U.S.C. 156(a)(5)(A)

The commercial marketing or use of the approved product after the regulatory review period is the first permitted commercial marketing or use of the approved product under the provisions of the Federal Food, Drug, and Cosmetic Act (21 U.S.C. 355) under which such regulatory review period occurred.

The length of extension of the patent term of U.S. Patent 4,312,860 claimed by Applicant is 1029 days. As noted in paragraph (10) above, IND 26,298 became effective on May 31, 1985; and NDA 20-044 was submitted February 16, 1990 and approved August 2, 1990. The 1029 day period is calculated by

adding one-half of the portion of the regulatory review period for the approved product beginning May 31, 1985 (i.e., the date on which IND 26,298 became effective) and ending February 16,1990 (i.e., the date on which NDA 20-044 for EXOSURF® was submitted) – 861 days – and all of the portion of the regulatory review period beginning February 16, 1990 and ending August 2, 1990 (i.e., the date on which NDA 20-044 for EXOSURF® was approved) – 168 days, for a total of 1029 days.

(13) Wellcome, as special agent for the Applicant, acknowledges a duty to disclose to the Commissioner of Patents and Trademarks and the Secretary of Health and Human Services any information which is material to any determinations to be made relative to the application for extension.

Attached hereto is a Declaration signed on behalf of the Applicant which meets the criteria set forth in 37 CFR 1.740 (17).

A check for $600 payable to the Commissioner of Patents and Trademarks is attached to cover the fee for this application for extension of term. In the event the actual fee differs from that specified above, it is requested that the overpayment be charged or the underpayment credited as authorized in the attached letter from Dr. Lawrence A. Nielsen.

Respectfully submitted,

Burroughs Wellcome Co., as Special Agent for
The Regents of the University of California
Alameda, California

by David A. Yeowell

David A. Yeowell
Vice President - Technical Development

LAN/jw*/10-USP 4,312,860

CERTIFICATION

The undersigned hereby certifies that this Application For Extension of Patent Term Under 35 U.S.C. 156 including its EXHIBITS and supporting papers is being submitted as duplicate originals.

9/27/90
Date

David A. Yeowell
David A. Yeowell

LAN/jw*/11-USP 4,312,860

Reflux: Reflux of Exosurf Neonatal into the endotracheal tube during dosing has been observed and may be associated with rapid drug administration. If reflux occurs, drug administration should be halted and, if necessary, peak inspiratory pressure on the ventilator should be increased by 4 to 5 cm H_2O until the endotracheal tube clears.

>20% Drop in Transcutaneous Oxygen Saturation: If transcutaneous oxygen saturation declines during dosing, drug administration should be halted and, if necessary, peak inspiratory pressure on the ventilator should be increased by 4 to 5 cm H_2O for 1 to 2 minutes. In addition, increases of FiO_2 may be required for 1 to 2 minutes.

Mucous Plugs: See WARNINGS.

OVERDOSAGE: There have been no reports of massive overdosage with Exosurf Neonatal.

DOSAGE AND ADMINISTRATION:

Preparation of Suspension: Exosurf Neonatal is best reconstituted immediately before use because it does not contain antibacterial preservatives. However, the reconstituted suspension is chemically and physically stable and remains sterile (when reconstituted using aseptic techniques) when stored at 2° to 30°C (36° to 86°F) for up to 12 hours following reconstitution.

Solutions containing buffers or preservatives should not be used for reconstitution. **Do Not Use Bacteriostatic Water for Injection, USP.** Each vial of Exosurf Neonatal should be reconstituted only with **8 mL** of the accompanying diluent (preservative-free Sterile Water for Injection) as follows:

1. Fill a 10 mL or 12 mL syringe with 8 mL preservative-free Sterile Water for Injection using an 18 or 19 gauge needle;
2. Allow the vacuum in the vial to draw the sterile water into the vial;
3. Aspirate as much as possible of the 8 mL out of the vial into the syringe (while maintaining the vacuum), then SUDDENLY release the syringe plunger.

Step 3 should be repeated three or four times to assure adequate mixing of the vial contents. If vacuum is not present, the vial of Exosurf Neonatal should not be used.

The appropriate dosage volume for the entire dose (5 mL/kg) should then be drawn into the syringe from **below** the froth in the vial (again maintaining the vacuum). If the infant weighs less than 1600 grams, unused Exosurf Neonatal suspension will remain in the vial after the entire dose is drawn into the syringe. If the infant weighs more than 1600 grams, at least two vials will be required for each dose.

Reconstituted Exosurf Neonatal is a milky white suspension with a total volume of 8 mL per vial. Each mL of reconstituted Exosurf Neonatal contains 13.5 mg colfosceril palmitate, 1.5 mg cetyl alcohol, 1 mg tyloxapol, and sodium chloride to provide a 0.1 N concentration. If the suspension appears to separate, gently shake or swirl the vial to resuspend the preparation. The reconstituted product should be inspected visually for homogeneity immediately before administration; if persistent large flakes or particulates are present, the vial should not be used.

Dosage: Accurate determination of weight at birth is the key to accurate dosing.

Prophylactic Treatment: The first dose of Exosurf Neonatal should be administered as a single 5 mL/kg dose as soon as possible after birth. Second and third doses should be administered approximately 12 and 24 hours later to all infants who remain on mechanical ventilation at those times.

Rescue Treatment: Exosurf Neonatal should be administered in two 5 mL/kg doses. The initial dose should be administered as soon as possible after the diagnosis of RDS is confirmed. The second dose should be administered approximately 12 hours following the first dose, provided the infant remains on mechanical ventilation. A small number of infants with RDS have received more than two doses of Exosurf Neonatal as rescue treatment. Definitive data on the safety and efficacy of these additional doses are not available (see PRECAUTIONS).

Use of Special Endotracheal Tube Adapter: With each vial of Exosurf Neonatal for Intratracheal Suspension, five different sized endotracheal tube adapters each with a special right angle Luer®-lock sideport are supplied. The adapters are clean but not sterile. The adapters should be used as follows:

1. Select an adapter size which corresponds to the inside diameter of the endotracheal tube.
2. Insert the adapter into the endotracheal tube with a firm push-twist motion.
3. Connect the breathing circuit wye to the adapter.
4. Remove the cap from the sideport on the adapter. Attach the syringe containing drug to the sideport.
5. After completion of dosing, remove the syringe and RECAP THE SIDEPORT.

Administration: The infant should be suctioned prior to administration of Exosurf Neonatal.

Exosurf Neonatal suspension is administered via the sideport on the special endotracheal tube adapter **WITHOUT INTERRUPTING MECHANICAL VENTILATION.**

Each Exosurf Neonatal dose is administered in two 2.5 mL/kg half-doses. Each half-dose is instilled slowly over 1 to 2 minutes (30 to 50 mechanical breaths) in small bursts timed with inspiration. After the first 2.5 mL/kg half-dose is administered in the midline position, the infant's head and torso are turned 45° to the **right** for 30 seconds while mechanical ventilation is continued. After the infant is returned to the midline position, the second 2.5 mL/kg half-dose is given in an identical fashion over another 1 to 2 minutes. The infant's head and torso are then turned 45° to the **left** for 30 seconds while mechanical ventilation is continued, and the infant is then turned back to the midline position. These maneuvers allow gravity to assist in the distribution of Exosurf Neonatal in the lungs.

During dosing, heart rate, color, chest expansion, facial expressions, the oximeter, and the endotracheal tube patency and position should be monitored. If heart rate slows, the infant becomes dusky or agitated, transcutaneous oxygen saturation falls more than 15%, or Exosurf Neonatal backs up in the endotracheal tube, dosing should be slowed or halted and, if necessary, the peak inspiratory pressure, ventilator rate, and/or FiO_2 turned up. On the other hand, rapid improvements in lung function may require immediate reductions in peak inspiratory pressure, ventilator rate, and/or FiO_2. (See WARNINGS and see below for additional information concerning administration.)

Suctioning should not be performed for two hours after Exosurf Neonatal is administered, except when dictated by clinical necessity.

General Guidelines for Administration: Administration of Exosurf Neonatal should not take precedence over clinical assessment and stabilization of critically ill infants.

Intubation: Prior to dosing with Exosurf Neonatal, it is important to ensure that the endotracheal tube tip is in the trachea and not in the esophagus or right or left mainstem bronchus. Brisk and symmetrical chest movement with each mechanical inspiration should be confirmed prior to dosing, as should equal breath sounds in the two axillae. In prophylactic treatment, dosing with Exosurf Neonatal need not be delayed for radiographic confirmation of the endotracheal tube tip position. In rescue treatment, bedside confirmation of endotracheal tube tip position is usually sufficient, if at least one chest radiograph subsequent to the last intubation confirmed proper position of the endotracheal tube tip. Some lung areas will remain undosed if the endotracheal tube tip is too low.

Monitoring: Continuous ECG and transcutaneous oxygen saturation monitoring during dosing are essential. In most infants treated prophylactically, it should be possible to initiate such monitoring prior to administration of the first dose of Exosurf Neonatal. For subsequent prophylactic and all rescue doses, arterial blood pressure monitoring during dosing is also highly desirable. After both prophylactic and rescue dosing, frequent arterial blood gas sampling is required to prevent post-dosing hyperoxia and hypocarbia (see WARNINGS).

Ventilatory Support During Dosing: The 5 mL/kg dosage volume may cause transient impairment of gas exchange by physical blockage of the airway, particularly in infants on low ventilator settings. As a result, infants may exhibit a drop in oxygen saturation during dosing, especially if they are on low ventilator settings prior to dosing. These transient effects are easily overcome by increasing peak inspiratory pressure on the ventilator by 4 to 5 cm H_2O for 1 to 2 minutes during dosing. FiO_2 can also be increased if necessary. In infants who are particularly fragile or reactive to external stimuli, increasing peak inspiratory pressure by 4 to 5 cm H_2O and/or FiO_2 20% just prior to dosing may minimize any transient deterioration in oxygenation. However, in virtually all cases it should be possible to return the infant to pre-dose settings within a very short time of dose completion.

Post-Dosing: At the end of dosing, position of the endotracheal tube should be confirmed by listening for equal breath sounds in the two axillae. Attention should be paid to chest expansion, color, transcutaneous saturation, and arterial blood gases. Some infants who receive Exosurf Neonatal and other surfactants respond with rapid improvements in pulmonary compliance, minute ventilation, and gas exchange (see WARNINGS). Constant bedside attention of an experienced clinician for at least 30 minutes after dosing is essential. Frequent blood gas sampling also is absolutely essential. Rapid changes in lung function require immediate changes in peak inspiratory pressure, ventilator rate, and/or FiO_2.

HOW SUPPLIED: Exosurf Neonatal for Intratracheal Suspension is supplied in a carton containing one 10 mL vial of Exosurf Neonatal for Intratracheal Suspension, one 10 mL vial of Sterile Water for Injection, and five endotracheal tube adapters (2.5, 3.0, 3.5, 4.0, and 4.5 mm I.D.). (NDC 0081-0207-01)

Store Exosurf Neonatal for Intratracheal Suspension at 15° to 30°C (59° to 86°F) in a dry place.

EDUCATIONAL MATERIAL: A videotape on dosing is available from your Burroughs Wellcome Co. representative. This videotape demonstrates techniques for safe administration of Exosurf Neonatal and should be viewed by health care professionals who will administer the drug.

Licensed under U.S. Patent Nos. 4312860 and 4826821

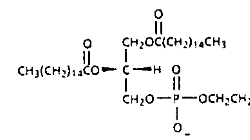
Wellcome

EXOSURF® Neonatal™
(COLFOSCERIL PALMITATE, CETYL ALCOHOL, TYLOXAPOL)
For Intratracheal Suspension

DESCRIPTION: Exosurf Neonatal (colfosceril palmitate, cetyl alcohol, tyloxapol) for Intratracheal Suspension is a protein-free synthetic lung surfactant stored under vacuum as a sterile lyophilized powder. Exosurf Neonatal is reconstituted with preservative-free Sterile Water for Injection prior to administration by intratracheal instillation. Each 10 mL vial contains 108 mg colfosceril palmitate, commonly known as dipalmitoylphosphatidylcholine (DPPC), 12 mg cetyl alcohol, 8 mg tyloxapol, and 47 mg sodium chloride. Sodium hydroxide or hydrochloric acid may have been added to adjust pH. When reconstituted with 8 mL Sterile Water for Injection, the Exosurf Neonatal suspension contains 13.5 mg/mL colfosceril palmitate, 1.5 mg/mL cetyl alcohol, and 1 mg/mL tyloxapol in 0.1 N NaCl. The suspension appears milky white with a pH of 5 to 7 and an osmolality of 185 mOsm/L.

The chemical names and structural formulas of the components of Exosurf Neonatal are as follows:

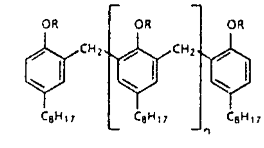
colfosceril palmitate
(1,2-dipalmitoyl-*sn*-3-phosphoglycerocholine)



cetyl alcohol
(1-hexadecanol)

$CH_3(CH_2)_{14}CH_2OH$

tyloxapol
(formaldehyde polymer with oxirane and 4-(1,1,3,3-tetramethylbutyl)phenol)



[R is $CH_2CH_2O(CH_2CH_2O)_mCH_2CH_2OH$;
m is 6 to 8; n is not more than 5]

CLINICAL PHARMACOLOGY: Surfactant deficiency is an important factor in the development of the neonatal respiratory distress syndrome (RDS). Thus, surfactant replacement therapy early in the course of RDS should ameliorate the disease and improve symptoms. Natural surfactant, a combination of lipids and apoproteins, exhibits not only surface tension reducing properties (conferred by the lipids), but also rapid spreading and adsorption (conferred by the apoproteins). The major fraction of the lipid component of natural surfactant is DPPC, which comprises up to 70% of natural surfactant by weight.

Although DPPC reduces surface tension, DPPC alone is ineffective in RDS because DPPC spreads and adsorbs poorly. In Exosurf Neonatal, which is protein free, cetyl alcohol acts as the spreading agent for the DPPC on the air-fluid interface. Tyloxapol, a polymeric long-chain repeating alcohol, is a nonionic surfactant which acts to disperse both DPPC and cetyl alcohol. Sodium chloride is added to adjust osmolality.

Pharmacokinetics: Exosurf Neonatal is administered directly into the trachea. Human pharmacokinetic studies of the absorption, biotransformation, and excretion of the components of Exosurf Neonatal have not been performed. Nonclinical studies, however, have shown that DPPC can be absorbed from the alveolus into lung tissue where it can be catabolized extensively and reutilized for further phospholipid synthesis and secretion. In the developing rabbit, 90% of alveolar phospholipids are recycled. In premature rabbits, the alveolar half-life of intratracheally administered H^3-labeled phosphatidylcholine is approximately 12 hours.

Animal Studies: In animal models of RDS, treatment with Exosurf Neonatal significantly improved lung volume, compliance and gas exchange in premature rabbits and lambs. The amount and distribution of lung water were not affected by Exosurf Neonatal treatment of premature rabbit pups. The extent of lung injury in premature rabbit pups undergoing mechanical ventilation was reduced significantly by Exosurf Neonatal treatment. In premature lambs, neither systemic blood flow nor flow through the ductus arteriosus were affected by Exosurf Neonatal treatment. Survival was significantly better in both premature rabbits and premature lambs treated with Exosurf Neonatal.

Clinical Studies: Exosurf Neonatal has been studied in the U.S. and Canada in controlled clinical trials involving more than 4400 infants. Over 10,000 infants have received Exosurf Neonatal through an open, uncontrolled, North American study designed to provide the drug to premature infants who might benefit and to obtain additional safety information (Exosurf Neonatal Treatment IND).

Prophylactic Treatment: The efficacy of a single dose of Exosurf Neonatal in prophylactic treatment of infants at risk of developing respiratory distress syndrome (RDS) was examined in three double-blind, placebo-controlled studies, one involving 215 infants weighing 500 to 700 grams, one involving 385 infants weighing 700 to 1350 grams, and one involving 446 infants weighing 700 to 1100 grams. The infants were intubated and placed on mechanical ventilation, and received 5 mL/kg Exosurf Neonatal or placebo (air) within 30 minutes of birth.

The efficacy of one versus three doses of Exosurf Neonatal in prophylactic treatment of infants at risk of developing RDS was examined in a double-blind, placebo-controlled study of 823 infants weighing 700 to 1100 grams. The infants were intubated and placed on mechanical ventilation, and received a first 5 mL/kg dose of Exosurf Neonatal within 30 minutes. Repeat 5 mL/kg doses of Exosurf Neonatal or placebo (air) were given to all infants who remained on mechanical ventilation at approximately 12 and 24 hours of age. An initial analysis of 716 infants is available.

The major efficacy parameters from these studies are presented in Table 1.

Table 1
Efficacy Assessments—Prophylactic Treatment

Number of Doses:	Single Dose		Single Dose		Single Dose		One Versus Three Doses	
Birth Weight Range:	500 to 700 grams		700 to 1350 grams		700 to 1100 grams		700 to 1100 grams	
Treatment Group	Placebo (Air)	EXOSURF	Placebo (Air)	EXOSURF	Placebo (Air)	EXOSURF	One EXOSURF Dose	Three EXOSURF Doses
Number of Infants:	N=106	N=109	N=185	N=176	N=222	N=224	N=356	N=360
	% of Infants		% of Infants		% of Infants		% of Infants	
Death ≤ Day 28[a]	53	50	11	6	21	15	16	9*
Death through 1 Year[a]	59	60	14	11	30	20**	17	12*
Death from RDS[b]	25	15*	4	3	10	5§	3	2
Intact Cardiopulmonary Survival[a,c]	29	25	69	78*	65	68	74	78
Bronchopulmonary Dysplasia (BPD)[a,d]	45	44	13	18	19	21	8	12
RDS Incidence[b]	73	71	16	12	55	55	63	58

[a] "Intent-to-treat" analyses (as randomized) except in the 700 to 1350 gram single dose study in which infants with congenital infections and anomalies were excluded — * p<0.05; ** p<0.01; § p=0.051
[b] "As treated" analyses
[c] Defined by survival through 28 days of life without bronchopulmonary dysplasia
[d] Defined by a combination of clinical and radiographic criteria

Rescue Treatment: The efficacy of Exosurf Neonatal in the rescue treatment of infants with RDS was examined in two double-blind, placebo-controlled studies. One study enrolled [illegible] infants weighing [illegible] grams; the second enrolled [illegible] infants weighing [illegible] grams and above. In the [illegible] studies, infants received an initial dose (5 mL/kg) of Exosurf Neonatal or placebo [illegible]

Table 2
Efficacy Assessments—Rescue Treatment

Number of Doses: / Birth Weight Range:	Two Doses 700 to 1350 grams		Two Doses 1250 grams and above	
Treatment Group: / Number of Infants:	Placebo (Air) N=213	EXOSURF N=206	Placebo (Air) N=623	EXOSURF N=614
	% of Infants		% of Infants	
Death ≤ Day 28[a]	23	11***	7	4*
Death through 1 Year[a]	27	15***	9	6§
Death from RDS[b]	10	3**	3	1*
Intact Cardiopulmonary Survival[b,c]	62	75**	88	93**
Bronchopulmonary Dysplasia (BPD)[a,d]	18	15	6	3*

[a] "Intent-to-treat" analyses (as randomized)
[b] "As-treated" analyses
[c] Defined by survival through 28 days of life without bronchopulmonary dysplasia
[d] Defined by a combination of clinical and radiographic criteria

* p<0.05
** p<0.01
*** p<0.001
§ p=0.067

Clinical Results: In these six controlled clinical studies, infants in the Exosurf Neonatal group showed significant improvements in FiO_2 and ventilator settings which persisted for at least 7 days. Pulmonary air leaks were significantly reduced in each study. Five of these studies also showed a significant reduction in death from RDS. Further, overall mortality was reduced for all infants weighing >700 grams. The one versus three-dose prophylactic treatment study in 700 to 1100 gram infants showed a further reduction in overall mortality with two additional doses.

Safety information is presented in Tables 3 and 4 (see ADVERSE REACTIONS). Beneficial effects in the Exosurf Neonatal group were observed for some safety assessments. Various forms of pulmonary air leak and use of pancuronium were reduced in infants receiving Exosurf Neonatal in all six studies.

Follow-up data at one year adjusted age are available on 1094 of 2470 surviving infants. Growth and development of infants who received Exosurf Neonatal in this sample were comparable to infants who received placebo.

INDICATIONS AND USAGE: Exosurf Neonatal is indicated for:

1. **Prophylactic** treatment of infants with birth weights of less than 1350 grams who are at risk of developing RDS (see PRECAUTIONS),
2. **Prophylactic** treatment of infants with birth weights greater than 1350 grams who have evidence of pulmonary immaturity, and
3. **Rescue** treatment of infants who have developed RDS.

For **prophylactic** treatment, the first dose of Exosurf Neonatal should be administered as soon as possible after birth (see DOSAGE AND ADMINISTRATION: General Guidelines for Administration).

Infants considered as candidates for **rescue** treatment with Exosurf Neonatal should be on mechanical ventilation and have a diagnosis of RDS by both of the following criteria:

1. Respiratory distress not attributable to causes other than RDS, based on clinical and laboratory assessments.
2. Chest radiographic findings consistent with the diagnosis of RDS.

During the clinical development of Exosurf Neonatal, all infants who received the drug were intubated and on mechanical ventilation. For three-dose prophylactic treatment with Exosurf Neonatal, the first dose of drug was administered as soon as possible after birth and repeat doses were given at approximately 12 and 24 hours after birth if infants remained on mechanical ventilation at those times. For rescue treatment, two doses were given; one between 2 and 24 hours of life, and a second approximately 12 hours later if infants remained on mechanical ventilation. Infants who received rescue treatment with Exosurf Neonatal had a documented arterial to alveolar oxygen tension ratio (a/A) <0.22.

CONTRAINDICATIONS: There are no known contraindications to treatment with Exosurf Neonatal.

WARNINGS:

Intratracheal Administration Only: Exosurf Neonatal should be administered only by instillation into the trachea (see DOSAGE AND ADMINISTRATION).

General:

The use of Exosurf Neonatal requires expert clinical care by experienced neonatologists and other clinicians who are accomplished at neonatal intubation and ventilatory management. Adequate personnel, facilities, equipment, and medications are required to optimize perinatal outcome in premature infants.

Instillation of Exosurf Neonatal should be performed **only** by trained medical personnel experienced in airway and clinical management of unstable premature infants. Vigilant clinical attention should be given to all infants prior to, during, and after administration of Exosurf Neonatal.

Acute Effects: Exosurf Neonatal can rapidly affect oxygenation and lung compliance.

Lung Compliance: If chest expansion improves substantially after dosing, peak ventilator inspiratory pressures should be reduced immediately, without waiting for confirmation of respiratory improvement by blood gas assessment. Failure to reduce inspiratory ventilator pressures rapidly in such instances can result in lung overdistention and fatal pulmonary air leak.

Hyperoxia: If the infant becomes pink and transcutaneous oxygen saturation is in excess of 95%, FiO_2 should be reduced in small but repeated steps (until saturation is 90 to 95%) without waiting for confirmation of elevated arterial pO_2 by blood gas assessment. Failure to reduce FiO_2 in such instances can result in hyperoxia.

Hypocarbia: If arterial or transcutaneous CO_2 measurements are <30 torr, the ventilator rate should be reduced at once. Failure to reduce ventilator rates in such instances can result in marked hypocarbia, which is known to reduce brain blood flow.

Pulmonary Hemorrhage: In the single study conducted in infants weighing <700 grams at birth, the incidence of pulmonary hemorrhage (10% vs 2% in the placebo group) was significantly increased in the Exosurf Neonatal group. None of the five studies involving infants with birth weights >700 grams showed a significant increase in pulmonary hemorrhage in the Exosurf Neonatal group. In a cross-study analysis of these five studies, pulmonary hemorrhage was reported for 1% (14/1420) of infants in the placebo group and 2% (27/1411) of infants in the Exosurf Neonatal group. Fatal pulmonary hemorrhage occurred in three infants; two in the Exosurf Neonatal group and one in the placebo group. Mortality from all causes among infants who developed pulmonary hemorrhage was 43% in the placebo group and 37% in the Exosurf Neonatal group.

Pulmonary hemorrhage in both Exosurf Neonatal and placebo infants was more frequent in infants who were younger, smaller, male, or who had a patent ductus arteriosus. Pulmonary hemorrhage typically occurred in the first 2 days of life in both treatment groups.

In more than 7700 infants in the open, uncontrolled study, pulmonary hemorrhage was reported in 4%, but fatal pulmonary hemorrhage was reported rarely (0.4%).

In the controlled clinical studies, Exosurf Neonatal treated infants who received steroids more than 24 hours prior to delivery or indomethacin postnatally had a lower rate of pulmonary hemorrhage than other Exosurf Neonatal treated infants. Attention should be paid to early and aggressive diagnosis and treatment (unless contraindicated) of patent ductus arteriosus during the first 2 days of life (while the ductus arteriosus is often clinically silent). Other potentially protective measures include attempting to decrease FiO_2 preferentially over ventilator pressures during the first 24 to 48 hours after dosing, and attempting to decrease PEEP minimally for at least 48 hours after dosing.

Mucous Plugs: Infants whose ventilation becomes markedly impaired during or shortly after dosing may have mucous plugging of the endotracheal tube, particularly if pulmonary secretions were prominent prior to drug administration. Suctioning of all infants prior to dosing may lessen the chance of mucous plugs obstructing the endotracheal tube. If endotracheal tube obstruction from such plugs is suspected, and suctioning is unsuccessful in removing the obstruction, the blocked endotracheal tube should be replaced immediately.

PRECAUTIONS:

General: In the controlled clinical studies, infants known prenatally or postnatally to have major congenital anomalies, or who were suspected of having congenital infection, were excluded from entry. However, these disorders cannot be recognized early in life in all cases, and a few infants with these conditions were entered. The benefits of Exosurf Neonatal in the affected infants who received drug appeared to be similar to the benefits observed in infants without anomalies or occult infection.

Prophylactic Treatment–Infants <700 Grams: In infants weighing 500 to 700 grams, a single prophylactic dose of Exosurf Neonatal significantly: improved FiO_2 and ventilator settings, reduced pneumothorax, and reduced death from RDS, but increased pulmonary hemorrhage (see WARNINGS). Overall mortality did not differ significantly between the placebo and Exosurf Neonatal groups (see Table 1). Data on multiple doses in infants in this weight class are not yet available. Accordingly, clinicians should carefully evaluate the potential risks and benefits of Exosurf Neonatal administration in these infants.

Rescue Treatment–Number of Doses: A small number of infants with RDS have received more than two doses of Exosurf Neonatal as rescue treatment. Definitive data on the safety and efficacy of these additional doses are not available.

Carcinogenesis, Mutagenesis, Impairment of Fertility: Exosurf Neonatal at concentrations up to 10,000 µg/plate was not mutagenic in the Ames Salmonella assay.

Long-term studies have not been performed in animals to evaluate the carcinogenic potential of Exosurf Neonatal.

The effects of Exosurf Neonatal on fertility have not been studied.

ADVERSE REACTIONS:

General: Premature birth is associated with a high incidence of morbidity and mortality. Despite significant reductions in overall mortality associated with Exosurf Neonatal, some infants who received Exosurf Neonatal developed severe complications and either survived with permanent handicaps or died.

In controlled clinical studies evaluating the safety and efficacy of Exosurf Neonatal, numerous safety assessments were made. In infants receiving Exosurf Neonatal, pulmonary hemorrhage, apnea and use of methylxanthines were increased. A number of other adverse events were significantly reduced in the Exosurf Neonatal group, particularly various forms of pulmonary air leak and use of pancuronium. (See CLINICAL PHARMACOLOGY: Clinical

Table 3
Safety Assessments[a]—Prophylactic Treatment

Number of Doses: / Birth Weight Range:	Single Dose 500 to 700 grams		Single Dose 700 to 1350 grams		Single Dose 700 to 1100 grams		One Versus Three Doses 700 to 1100 grams	
Treatment Group: / Number of Infants:	Placebo (Air) N=108	EXOSURF N=107	Placebo (Air) N=193	EXOSURF N=192	Placebo (Air) N=222	EXOSURF N=224	One EXOSURF Dose N=356	Three EXOSURF Doses N=360
	% of Infants		% of Infants		% of Infants		% of Infants	
Intraventricular Hemorrhage (IVH)								
Overall	51	57	31	27	36	36	38	35
Severe IVH	26	25	10	8	13	14	9	9
Pulmonary Air Leak (PAL)								
Overall	52	48	16	11	32	25	29	27
Pneumothorax	23	10*	5	6	19	11*	14	12
Pneumopericardium	1	4	2	0	<1	1	1	1
Pneumomediastinum	2	1	2	3	7	1**	3	2
Pulmonary Interstitial Emphysema	43	44	13	7*	26	20	23	22
Death from PAL	4	6	<1	<1	2	1	2	1
Patent Ductus Arteriosus	49	53	66	70	50	55	59	57
Necrotizing Enterocolitis	2	4	11	13	3	4	6	2*
Pulmonary Hemorrhage	2	10**	2	4	1	4	4	6
Congenital Pneumonia	4	4	2	4	2	2	1	1
Nosocomial Pneumonia	10	10	2	4	4	7	14	15
Non-Pulmonary Infections	33	35	34	39	28	29	35	34
Sepsis	30	34	30	34	23	24	30	27
Death From Sepsis	4	4	3	3	1	2	3	2
Meningitis	4	6	3	1	2	3	1	2
Other Infections	7	4	5	3	6	10	10	11
Major Anomalies	3	1	2	4	7	4	4	4
Hypotension	70	77	52	47	59	62	54	50
Hyperbilirubinemia	22	21	63	61	27	31	20	21
Exchange Transfusion	4	3	1	2	2	2	3	1
Thrombocytopenia[b]	21	25	not available		9	8	12	10
Persistent Fetal Circulation	0	1	1	1	0	2*	1	<1
Seizures	11	8	2	2	11	9	6	5
Apnea	34	33	76	73	55	65*	62	68
Drug Therapy								
Antibiotics	96	99	98	96	98	99	>99	99
Diuretics	55	60	39	37	59	63	64	65
Anticonvulsants	14	18	23	24	20	16	9	8
Inotropes	46	40	20	20	26	20	28	27
Sedatives	62	71	65	64	63	57	52	52
Pancuronium	19	11	22	14*	19	13*	15	11
Methylxanthines	38	43	77	77	61	72*	75	82*

[a] All parameters were examined with "as-treated" analyses.
[b] Thrombocytopenia requiring platelet transfusion.

* p<0.05
** p<0.01

Table 4
Safety Assessments[a]—Rescue Treatment

Number of Doses: / Birth Weight Range:	Two Doses 700 to 1350 grams		Two Doses 1250 grams and above	
Treatment Group: / Number of Infants:	Placebo (Air) N=213	EXOSURF N=206	Placebo (Air) N=622	EXOSURF N=615
	% of Infants		% of Infants	
Intraventricular Hemorrhage (IVH)				
Overall	48	52	23	18*
Severe IVH	13	9	5	4
Pulmonary Air Leak (PAL)				
Overall	54	34***	30	18***
Pneumothorax	29	20*	20	10***
Pneumopericardium	4	1	1	2
Pneumomediastinum	8	4	5	2**
Pulmonary Interstitial Emphysema	48	25***	24	13***
Death from PAL	7	3	<1	1
Patent Ductus Arteriosus	66	57	54	45*
Necrotizing Enterocolitis	3	3	1	2
Pulmonary Hemorrhage	3	1	<1	1
Congenital Pneumonia	2	3	2	2
Nosocomial Pneumonia	5	7	2	2
Non-Pulmonary Infections	19	22	13	13
Sepsis	15	17	8	8
Death From Sepsis	<1	<1	1	<1
Meningitis	1	<1	1	<1*
Other Infections	5	8	5	6
Major Anomalies	3	3	4	4
Hypotension	62	57	50	39**
Hyperbilirubinemia	17	19	12	10
Exchange Transfusion	3	4	1	2
Thrombocytopenia[b]	10	11	4	<1**
Persistent Fetal Circulation	1	1	6	2**
Seizures	10	10	6	3*
Apnea	48	65**	37	44*
Drug Therapy				
Antibiotics	100	99	98	98
Diuretics	60	65	45	34***
Anticonvulsants	17	17	10	5**
Inotropes	36	31	27	16***
Sedatives	72	68	76	64***
Pancuronium	34	17**	33	15***
Methylxanthines	62	74**	49	53

[a] All parameters were examined with "as-treated" analyses.
[b] Thrombocytopenia requiring platelet transfusion.

* p<0.05
** p<0.01
*** p<0.001

Pulmonary Hemorrhage: See WARNINGS.

Abnormal Laboratory Values: Abnormal laboratory values are common in critically ill, mechanically ventilated, premature infants. A higher incidence of abnormal laboratory values in the Exosurf Neonatal group was not reported.

Events During Dosing: Data on events during dosing are available from more than 8800 infants in the open, uncontrolled clinical study (Table 5).

Table 5
Events During Dosing in the Open, Uncontrolled Study[a]

Treatment Type: / Number of Infants:	Prophylactic Treatment N=1127	Rescue Treatment N=7711
	% of Infants	% of Infants
Reflux of Exosurf	20	11
Drop in O_2 saturation (≥ 20%)	6	22
Rise in O_2 saturation (≥ 10%)	5	6
Drop in transcutaneous pO_2 (≥ 20 mm Hg)	1	8
Rise in transcutaneous pO_2 (≥ 20 mm Hg)	2	5
Drop in transcutaneous pCO_2 (≥ 20 mm Hg)	<1	1
Rise in transcutaneous pCO_2 (≥ 20 mm Hg)	1	3
Bradycardia (<60 beats/min)	1	3
Tachycardia (>200 beats/min)	<1	<1
Gagging	1	2
Mucous Plugs	<1	<1

[a] Infants may have experienced more than one event.
Investigators were prohibited from adjusting FiO_2 and/or ventilator settings during dosing unless significant clinical deterioration occurred.

EXHIBIT 1

SPECIAL POWER OF ATTORNEY

Know All Men By These Presents, that The Regents of the University of California, organized under the laws of California and having a place of business at 1320 Harbor Bay Parkway, Suite 150, Alameda, California 94501, do hereby make, constitute and appoint Burroughs Wellcome Co., a corporation organized under the laws of the State of North Carolina and having its principal place of business at 3030 Cornwallis Road, Research Triangle Park, North Carolina 27709, as their special, true and lawful agent and attorney for the sole and limited purpose of preparing and filing with the U. S. Patent and Trademark Office a Patent Term Extension Application pursuant to 35 U.S.C. 156 in respect of U. S. Patent No. 4,312,860, which Patent is owned by The Regents of the University of California, and prosecuting said Application; and to do and perform each and every act in connection with the above-stated purpose which Burroughs Wellcome Co. deems necessary or desirable.

IN WITNESS WHEREOF, The Regents of the University of California have caused this instrument to be executed by their duly authorized officer, and their seal to be affixed hereto, as of the 21st day of September, 1990.

THE REGENTS OF THE
UNIVERSITY OF CALIFORNIA

By: W T Danis

Title: Asst Dir, PTCO

[Corporate Seal]

Attest: _____

Title: _____

STATE OF CALIFORNIA
COUNTY OF ALAMEDA ss.

On this 21 day of September, 1990, before me, LINDA S. STEVENSON, A Notary Public, State of California, duly commissioned and sworn, personally appeared William T. Davis personally known to me to be the Assistant Director, Patent, Trademark & Copyright Office on behalf of The Regents of the University of California, a public corporation, and known to me to be the person who executed the within instrument on behalf of said corporation and acknowledged to me that The Regents of the University of California executed the same.

OFFICIAL SEAL
LINDA S. STEVENSON
NOTARY PUBLIC-CALIFORNIA
ALAMEDA COUNTY
My Commission Expires March 12, 1991

IN WITNESS WHEREOF, I have hereunto set my hand and affixed my Official Seal, in the County of Alameda the day and year in this Certificate first above written.

Linda S. Stevenson
LINDA S. STEVENSON, Notary Public, State of California

My Commission Expires March 12, 1991

EXHIBIT 3

(To Application for Extension of Patent Term of U.S. Patent No. 4,312,860)

EXOSURF® Neonatal™ (colfosceril palmitate, cetyl alcohol, tyloxapol) for Intratrachael Suspension[1]

Chronology of Significant Activities During Regulatory Review Period[2]

Relating to IND 26, 298

05/01/85	Original IND submitted to FDA for Dr. Roderic H. Phibbs, M.D., University of California, San Francisco.
08/27/85	Letter from FDA to Dr. Phibbs regarding IND submission of 05/01/85; requesting additional manufacturing and controls data, pharmacological data and clarification regarding planned clinical studies.
10/24/85	Letter to FDA submitted by Dr. Phibbs in response to their letter of 08/27/85, regarding the pharmacological and clinical studies concerns.
12/04/85	Letter to FDA submitted by Dr. Phibbs in response to their letter of August 27, 1985, concerning the questions about manufacturing and controls.

Relating to IND 28,006

03/04/86	Submitted original IND providing for conduct of initial clinical study 01.
03/12/86	Letter from FDA acknowledging our original IND submission of 03/04/86 assigning number 28,006.
10/27/86	Minutes of meeting with FDA discussing clinical issues in the development of EXOSURF.
10/29/86	Amended IND to provide for clinical study 02, "Effects of EXOSURF Prophylaxis at Birth in High Risk Premature Infants: A Double-Blind, Randomized, Parallel Study," to be conducted by Anthony J. Corbet, M.D.

[1] In the IND and NDA submissions the product was named EXOSURF PEDIATRIC STERILE POWDER. The FDA was advised July 25, 1990 of the trade name change to EXOSURF® Neonatal™ (colfosceril palmitate, cetyl alcohol, tyloxapol) for Intratracheal Suspension.

[2] During the regulatory review period, in addition to the activities specifically described in this EXHIBIT 3, Burroughs Wellcome Co. had numerous additional communications with the FDA in the form of submissions, meetings and telephone calls relating to IND 28,006 and NDA 20-044.

11/15/86	Telephone call to FDA to discuss procedures for handling laboratory values and reporting deaths during EXOSURF development
02/09/87	Meeting with FDA to discuss computerized clinical data.
03/11/87	Letter from FDA with questions and comments concerning protocol 02, "Effects of EXOSURF Prophylaxis at Birth in High Risk Premature Infants: A Double-Blind, Randomized, Parallel Study".
03/12/87	Submitted progress report.
06/04/87	Letter to FDA responding to comments and requests in FDA letters of October 31, 1986 and March 11,1987, regarding protocol 02-01. Also, submitted minutes of February 9, 1987 meeting.
06/04/87	Informal meeting with FDA to discuss toxicity issues that had arisen during clinical studies with another surfactant under development.
06/10/87	Letter from FDA with comments regarding safety issues and requests for our input on animal toxicity studies.
06/22/87	Letter to FDA to confirm the meeting scheduled for June 22, 1987 to discuss preclinical studies with surfactants.
07/08/87	Letter from FDA with comments concerning our June 4, 1987 response to FDA concerns regarding study 02, "Effects of EXOSURF Prophylaxis at Birth in High Risk Premature Infants: A Double-Blind, Randomized, Parallel Study".
07/30/87	Amended IND to provide for clinical study 03, "U.S.Multicenter EXOSURF Tiny Baby Prophylaxis Trial: A Mortality Study", to be conducted by Philip Sunshine, M.D.
08/25/87	Letter from FDA with comments regarding our July 30, 1987 amendment to provide for clinical study 03, "U.S. Multicenter EXOSURF Tiny Baby Prophylaxis Trial: A Mortality Study".
09/18/87	Amended IND to provide for clinical study 04,"EXOSURF Prophylaxis and Intact Cardiopulmonary Survival in High Risk Premature Infants: A U.S.Multicenter Trial", to be conducted by Frans Walther, M.D., Ph.D., and Hakan Sundell, M.D.
09/23/87	Amended IND to provide for clinical study 05,"Effects of EXOSURF Rescue Treatment on Intact Cardiopulmonary Survival in Smaller Premature Infants with Respiratory Distress Syndrome: A U.S.Multicenter Trial", and clinical study 06, "Effects of EXOSURF Rescue Treatment on Morbidity in Larger Infants with Respiratory Distress Syndrome: A U.S.Multicenter Trial", to be conducted by Frans Walther, M.D., Ph.D.
10/22/87	Advisory Committee Meeting - presentation on EXOSURF.
10/22/87	Letter from FDA with comments concerning protocol 05, "Effects of EXOSURF Rescue Treatment on Intact Cardiopulmonary Survival in Smaller Premature Infants with Respiratory Distress Syndrome: A U.S.Multicenter Trial", and protocol 06, "Effects of EXOSURF Rescue Treatment on Morbidity in Larger Infants

	with Respiratory Distress Syndrome: A U.S.Multicenter Trial", submitted on September 23,1987.
10/28/87	Amended IND to provide for the following clinical trials to be conducted in Canada: Protocol 07 - "Canadian Multicenter EXOSURF Tiny Baby Salvage Trial: A Mortality Study." Protocol 08 - "Canadian Multicenter Trial: Effects of EXOSURF Rescue Treatment on Intact Cardiopulmonary Survival in Smaller Infants with Respiratory Distress Syndrome." Protocol 09 - "Effects of EXOSURF Rescue Treatment on Morbidity in Larger Infants with Respiratory Distress Syndrome: A Canadian Multicenter Trial."
12/22/87	Letter to FDA in response to their August 25, 1987 letter regarding clinical development plans.
01/26/88	Letter from FDA with comments regarding December 22, 1987 submission concerning protocol development.
03/02/88	Letter to FDA in response to their letter of October 22, 1987, concerning clinical endpoints.
10/06/88	Letter to FDA in response to their letter of April 1, 1988, concerning the use of "time on ventilator" as a primary outcome.
10/06/88	Submitted summary of the development of EXOSURF as requested in FDA telephone conversation of July 28, 1988.
02/21/89	Telephone call to FDA to discuss the preliminary results for the 04 study.
02/28/89	Amended IND to provide for Protocol 13 - "EXOSURF Pediatric Multiple Dose Prophylaxis Study in High Risk Premature Infants: A Multicenter Trial" to be conducted by David Easa, M.D., Jeffrey Gerdes,M.D., Matthew Sell, M.D., Donnal Walter, M.D., Michael LeBlanc, M.D., Martha Mullett, M.D.,Anthony Corbet, M.D., and Frans Walther, M.D.
04/27/89	Letter from FDA informing us that they had determined that EXOSURF may qualify for handling under the procedures delineated under Subpart E for expedited review and that if we wish to pursue development of this drug under the provisions of Subpart E, we must request this in writing.
04/28/89	Meeting with FDA to discuss the Treatment IND.
05/17/89	Meeting with FDA to discuss the manufacturing and controls requirements for the Treatment IND for EXOSURF Pediatric.
05/25/89	Submitted the Independent Advisory Panel's evaluation of the April, 1989 safety report
06/01 to 06/16/89	Meetings with FDA to discuss the EXOSURF Pediatric Treatment IND.
06/16/89	Meeting with FDA to discuss the EXOSURF Pediatric Treatment IND.
07/13/89	Letter to FDA requesting "E Designation" for EXOSURF in the treatment of neonatal respiratory distress syndrome.

08/24/89	Letter to FDA requesting a pre-NDA conference during the week of August 28, 1989 to discuss the initial NDA; re: preclinical and clinical data sections.
08/29/89	EXOSURF Pediatric Pre-NDA meeting held.
09/18/89	Letter from FDA informing us that our request of July 13, 1989 for "Subpart E Designation" of 21 CFR Part 312 qualifies for the special procedures designed to expedite the development, evaluation and marketing of new therapies.
09/29/89	Letter to FDA to confirm the meeting scheduled on October 5, 1989 to discuss clinical issues regarding the upcoming NDA for EXOSURF Pediatric Sterile.

Relating to NDA 20-044

12-12-89	As agreed, pre-submitted the Nonclinical Pharmacology and Toxicology Technical Section for the original NDA.
12-15-89 to 08-01-90	Manufacturing discussions between FDA and Burroughs Wellcome Co.
12-15-89	Pre-submitted the Chemistry, Manufacturing, and Controls Technical Section of the original NDA.
12-20-89	Pre-submitted the Human Pharmacokinetics and Bioavailability Technical Section for the original NDA.
02-02-90	Letter to FDA regarding environmental assessment requirements in an effort to expedite the review of pending export requests.
02-16-90	Submitted original NDA.
03-05-90	Letter from FDA acknowledging receipt of our original NDA submitted February 16, 1990; stating that the filing date will be April 17, 1990.
03-09-90	Official notification to FDA of Burroughs Wellcome Co.'s intent to exercise orphan drug exclusivity.
04-04-90	As requested by FDA, submitted a listing of IND's under which EXOSURF has been studied as an amendment to the Clinical and Statistical Sections of the NDA.
05-03-90	Submitted additional exploratory analysis of the original NDA data as requested by FDA in April 18, 1990 telephone conversation.
07-10-90	Meetings with FDA discussing: 1) the Environmental Assessment report; 2) the trade name; and 3) storage limitations.
07-11-90	Submitted a revised Environmental Assessment updated to include requested information contained in May 3, 1990 review draft letter.

07-16-90 to 08-17-90	Labeling negotiations between FDA and Burroughs Wellcome Co.
07-18-90	As agreed by FDA in July 17, 1990 telephone conversation, submitted a revised version of our July 11, 1990 Environmental Assessment.
07-23-90	As requested by FDA, submitted Summary Basis of Approval for the original NDA.
07-23-90	Pulmonary-Allergy Drugs Advisory Committee Meeting.
07-24-90	As requested, panafaxed to FDA patent and exclusivity information.
07-25-90	Advised FDA that the trade name will be changed to EXOSURF Neonatal for Intratracheal Suspension.
08-02-90	Letter from FDA approving the NDA.

EXHIBIT 2

United States Patent [19]

Clements

[54] **LUNG SURFACTANT COMPOSITIONS**

[75] Inventor: **John A. Clements**, Tiburon, Calif.

[73] Assignee: **Regents of the University of California**, Berkeley, Calif.

[21] Appl. No.: **200,216**

[22] Filed: **Oct. 24, 1980**

[51] **Int. Cl.**[3] **A01N 57/26**
[52] **U.S. Cl.** **424/199**
[58] **Field of Search** 424/199

[56] **References Cited**

U.S. PATENT DOCUMENTS

3,577,446	5/1971	Rakmit	424/199
4,129,650	12/1978	Betzing et al.	424/199

Primary Examiner—Elbert L. Roberts

Attorney, Agent, or Firm—Phillips, Moore, Weissenberger, Lempio & Majestic

[57] **ABSTRACT**

A synthetic protein-free lung surfactant composition is utilized to temporarily substitute for natural lung surfactant in the mammalian lung where such natural lung surfactant is absent or in low concentration. The synthetic surfactant composition consists essentially of a major amount of 1,2-dipalmitoyl-sn-3-glycerophosphoryl choline (DPPC), and a minor amount of a fatty alcohol, preferably a fatty alcohol having from 14 to 18 carbon atoms, and especially n-hexdecan-1-ol. The synthetic surfactant composition is administered directly into the lungs of a distressed subject to create a film on the alveolar interfacial surfaces and reduce surface tension. Expansion of the alveolar spaces is thereby facilitated.

9 Claims, No Drawings

LUNG SURFACTANT COMPOSITIONS

The invention described herein was made in the course of, or under, a grant from the National Institutes of Health.

DESCRIPTION

BACKGROUND OF THE INVENTION

The present invention is directed to compositions useful in alleviating the symptoms of mammalian respiratory distress syndrome (RDS) which may occur in the newborn, and especially in the prematurely newborn, as well as, in many instances in the adult when disease or functional difficulties bring about lung failure characterized by the deficiency of lung surfactant. The invention compositions may be introduced into the lungs of the distressed subject to temporarily provide the surfactant required for proper pulmonary function.

In the past several decades, the findings and writings of a number of investigators have brought greatly increased understanding in the medical community of the physiology of the mammalian lung; especially pertaining to the mechanisms involved in the transfer of gases from the air spaces in the lungs across the lining tissues to the underlying vascular system. These studies have revealed the critical role played by a liquid film which lines the tissue surfaces. This role is based upon basic physical principles which have been known for several hundred years, but whose application to the operation of the mammalian lung has only reached general recognition within the past 20 years or so.

Specifically, the basic physics principles involve the functioning of surface tension, i.e., the physical phenomenon exhibited by liquid surfaces brought about by intermolecular forces and resulting in a "skin like" effect. This phenomenon underlies the tendency of the lung's air sacs, or alveoli, to expell gas at all times during the respiratory cycle. If sufficiently low surface tension forces are not maintained at the air-lung tissue interface, the alveoli collapse during exhalation. Even the inspiration of air through the bronchi may be ineffective in inflating the collapsed alveoli and gas exchange into the pulmonary circulatory system may be inadequate.

Establishing and maintaining low surface tension at the alveolar surfaces is accomplished by an intricate biological system associated with alveolar lung tissue. Special cells, known as alveolar Type II, synthesize a complex mixture of lipids, proteins, glycerides and fatty acids. This complex is stored in the form of lamellar bodies within the alveolar Type II cells. By a mechanism little understood, the lamellar bodies are extruded from the alveolar Type II cells into alveolar lumen where the lamellae unwind and distribute the lipid, protein, glyceride, etc. molecules throughout the liquid film which bathes the entire cellular covering of the alveolar walls. These molecules, which may be generically referred to as "lung surfactant," migrate to the surface of the liquid film where they produce an essentially mono-molecular, all pervasive layer thereon.

The surfactant, effectively lowers the surface tension of the film to low values (circa 10 millineutons/meter) sufficient to maintain alveolar inflation during all phases of the respiratory cycle.

The chemical composition of "lung surfactant" has been investigated and the results have been published in a number of papers, e.g. Respiratory Distress Syn-

drome, *Academic Press Inc.*, 1973, pp. 77-98. Such studies indicate that natural lung surfactant is a complex mixture of many components of which the major component is a lipid, dipalmitoyl phosphatidyl choline (according to current naming criteria more correctly, 1,2-dipalmitoyl-sn-3-glycerophosphoryl choline). Dipalmitoyl phosphatidyl choline, commonly abbreviated as DPPC, occurs in lung surfactant to the extent of about 41% by weight. Mixed monenoic lecithins make up about 25% by weight; cholesterol makes up about 9% by weight; mixed proteins about 9% by weight; phosphatidyl ethanolamine, about 5%; various glycerides and phosphatidyl serine and phosphatidyl glycerol, about 4%, respectively; lysolecithin, about 2%; with sphingomyelin and fatty acids, each about 1%. The above noted materials and %'s are for surfactant removed from canine lungs; however, the mix of materials and %'s generally hold true for the higher mammals. For instance, both bovine and human lung surfactant also comprise a similar mix, with DPPC running in the same range of approximately 40% by weight.

Respiratory distress syndrome occurs when the necessary surfactant is either absent from, or is seriously depleted in, the liquid lining of the alveolar spaces. The most common occurrance is in the newborn and especially in the premature newborn, wherein development of the alveolar Type II cells has not yet arrived at a stage sufficient to generate the necessary surfactant material. The maturation of the alveolar Type II cells normally occurs within the last several weeks of full term gestation. However, in some instances congenital defects interfere with and/or delay maturation of the alveolar Type II cells; or more commonly in the instance of premature birth, maturation has not yet progressed sufficiently to generate the necessary surfactant.

In other instances, interruption of the generation of surfactant may occur in the mature and/or adult individual under the impact of disease and/or trauma.

It will be apparent from what has been noted herein and before that the lack of maturation of the surfactant generating mechanisms in the newborn and especially in the prematurely newborn, or the interruption of the surfactant generating mechanism resulting from disease or trauma, will result in the absence or the diminution of the necessary surfactant on the lining of the alveolar spaces. The absence of the necessary surfactant eliminates or may drastically interfer with the ability of the newborn lung to properly inflate as respiration begins. Similarly, collapse or deflation of the alveolar spaces occurs in the mature lung when the supply of surfactant is interrupted or diminished because of disease or trauma.

The absence or loss of lung surfactant is manifest by severe respiratory distress, which if not managed by medical intervention may most usually result in death. In the past, such medical intervention included such measures as supplying high levels of oxygen; positive pressure application to the lungs to provide adequate pulmonary ventilation; adequate attention to the maintenance of nutrition, fluid balance, blood volume, and blood pressure etc. In addition, in the case of the premature newborn it has been determined that the introduction of corticosteroids actively induces rapid maturation of the natural surfactant production system. Such steroid therapy, however, must be undertaken before the actual premature birth occurs in order to be truly effective in achieving early maturation of the surfactant

producing systems. With recent techniques of analyzing amniotic fluids, tests have been devised for determining the presence of adequate amounts of surfactant in the unborn fetus. Where it is anticipated that a premature birth will occur, such tests can be performed and if inadequate levels of surfactant are noted, steroid therapy can be instituted to hasten the maturation of the natural surfactant production systems.

Rather fortuitously soon after birth the corticosteroid systems begin and/or increase production of the corticosteroids internally and if the individual can be maintained for relatively short periods of time, in the matter of several days, maturation of the surfactant production systems will occur. Under these circumstances sufficient surfactants will soon be released into the alveolar surfaces to produce the low surface tension necessary to the full and unassisted expansion to maintain normal respiratory function.

Therefore, it becomes extremely critical to somehow manage the respiratory distress for a relatively short period of time (normally for a period of several days) until the natural systems can come into play and take over their role in maintaining a normal expansion of the alveolar spaces.

As pointed out above, in the past, management has included positive pressure pulmonary ventilation along with the monitoring and maintenance of secondary functions. However, with the discovery of the nature of lung surfactant, some work has been done to replace the lacking surfactant with exogenous surfactant components. Generally speaking, however, such attempts have been unsuccessful until Fujiwara and his coworkers used cow lung extract fortified with DPPC and phosphatidylglycerol, two of the principal components of natural lung surfactant. Fujiwara, et al. reported their work in Lancet 1:55, January 1980.

One of the possible shortcomings of a substitute surfactant derived from animal lung extracts are its undefined nature, the possibility of contamination with micro-organisms, and especially the presence of foreign proteins which may lead to possible sensitization in the individual to whom such extracts are administered. It is therefore desirable to develop a lung surfactant substitute whose composition is completely defined, whose production may essentially exclude any possibility of microbial contamination, and in which, antigenic proteins are completely absent.

With regard to the preparation of artificial lung surfactant compositions which are free of protein, I. L. Metcalfe and his coworkers have reported (J. Applied Physiology: Respiratory Environmental Exercise Physiology 49:34, 1980) that a composition of 70% DPPC, 20% egg phosphatidylcholine, 10% phosphotidylinositol and 1% palmitic acid, exhibits acceptable properties. Similarly, C. J. Morley at the 16th International Congress of Pediatrics held at Barcelona, September 1980 reported that an artificial surfactant consisting of DPPC and unsaturated phosphatidylglycerol shows promise.

Despite the reports of synthetic surfactant noted above, the preparation of a protein free synthetic lung surfactant substitute suitable as a temporary replacement for natural lung surfactant has been quite difficult since the physiochemical characteristics of natural lung surfactant are complex and at times contradictory. The principal characteristics of a lung surfactant are (1) it must absorb very rapidly from bulk phase to the liquid interface lining the alveolar tissues and spread a film

thereon. The film must be formed rapidly since newborns have a high respiratory rate and only a few tenths of a second is available during inspiration to form the film while the air spaces are expanding. (2) The surfactant surface film must be stable to ensure that the surface tension remains at a low value (not more than 10 mN/m) during expiration. The stable film ensures that as transpulmonary pressure falls, the alveolar spaces remain expanded and functional; and that residual volume does not decrease to zero. (3) Although some of the surfactant material inevitably is forced from the interfacial film during expiration, it is essential that the surfactant have sufficient mobility to reenter the interface during the next expansion. Such properties of the surfactant ensures that its loss from the interfacial film is not so high as to require excessive dosage volumes and/or rates.

Some of the requirements for the surfactants as noted above, appear to be contradictory insofar as the physicochemical properties of the lung surfactant materials are concerned. Thus, the high molecular mobility required for rapid adsorption and respreading into the interfacial film contradicts the low mobility necessary for a stable and persistent film. In natural lung surfactant, this contradiction is apparently resolved through the complexity of the multicomponents as noted above which are organized around a specific protein. Such complex material apparently has the ability to spontaneously undergo the necessary molecular sorting and phase changes required to satisfy these apparently contradictory physico chemical requirements. Thus the preparation of a simple, yet effective synthetic lung surfactant appears to be fought with difficulty.

BRIEF SUMMARY OF THE INVENTION

The present invention is broadly concerned with synthetic lung surfactant compositions and more specifically with simple, easily and inexpensively prepared surfactant compositions which are free from proteins, are made from known components securable from common industrial sources.

The synthetic lung surfactant compositions consist essentially of two components, more particularly, with synthetic lung surfactant compositions derived from mixtures of dipalmitoyl phosphatidylcholine and fatty alcohols. The dipalmitoyl phosphatidylcholine (DPPC) constitutes the major component of the surfactant composition while the fatty alcohol comprises a minor component thereof. The fatty alcohol component of the compositions may be any of a number of fatty alcohols having from 14–18 carbon atoms and may be either saturated or unsaturated. The much preferred fatty alcohol, however, is hexadecanol i.e. n-hexadecan-1-ol. Unsaturated fatty alcohols such as oleic alcohol may also be utilized in the surfactant compositions. Other fatty alcohols may also be utilized so long as they satisfy the criteria for the synthetic lung surfactant composition as noted above.

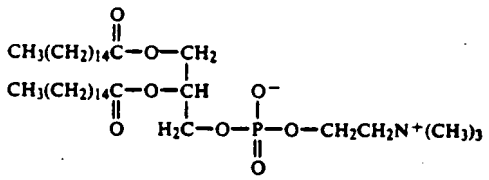
Suspensions of the synthetic lung surfactant are utilized for the treatment for respiratory distress syndrome in mammals by administering suspensions (aqueous or saline) of the surfactant directly into the lungs of the distressed subject.

Both DPPC and the fatty alcohol component are substances which occur naturally in mammalian tissues, although they do not occur together as a specific moiety. DPPC in fact occurs as the principal component in natural lung surfactant; however, the fatty alcohols of

the present composition are not known to occur naturally in lung tissue. Since both components of the surfactant composition do occur naturally within mammalian tissues, they are also metabolizable and their eventual elimination from a subject is accomplished by normal processes. Similarly, the hazard associated with the introduction into the organism of foreign substances is of no consideration with the present compositions.

DETAILED DESCRIPTION OF THE INVENTION

The synthetic lung surfactant compositions of this invention are protein-free and consist essentially of two components. The major component is dipalmitoyl phosphatidyl choline (DPPC), which is also the major component of naturally occurring lung surfactant. DPPC has been synthesized in the laboratory. It is a lipid, i.e., one of the broad class of organic compounds found in cells which are extractable by nonpolar solvents such as chloroform, ether, and benzene. It is comprised of two palmito-moieties linked to the phospho-glyceride moiety, phosphatidyl choline. The simple structural formula may be depicted as:



The lipid may be obtained in high purity on the commercial market.

The dipalmitoyl phosphatidyl choline is an essential component of the synthetic surfactant compositions and accounts for some of the desired properties of lung surfactant i.e., it forms very stable monolayers at 37° C., and is a principal component of natural lung surfactant. DPPC may be present in the synthetic compositions over a fairly wide range, although in any event as the major component. It has been tested at a percentage of as low as 82%, and as high as 94% by weight with no noted change in the surfactant's in vitro properties. Generally, however, DPPC is preferred in about 90% by weight in the surfactant composition.

The second component of the synthetic surfactant compositions is a fatty alcohol having carbons in the range of from about 14 to 18. Such fatty alcohols may be either saturated or unsaturated, although the saturated alcohol, hexadecanol (n-hexadecan-1-ol) is greatly preferred. The unsaturated alcohol, oleic alcohol, has also been combined with DPPC and the resultant surfactant appears to have the necessary properties.

Any of the closely related fatty alcohols in the C-14 to C-18 range can also be utilized so long as the resultant surfactant composition satisfies the required properties enumerated in the background section above.

The fatty alcohols are available in high purity on the commercial market. The alcohol component constitutes a minor portion of the surfactant composition, being present in an amount ranging from about 5 or 6% to about 18% or 20% by weight of the composition. The preferred composition of the synthetic surfactants of the invention is DPPC in about 90% by weight and hexadecanol in about 10% by weight. However, the percentages may be altered as noted above without unduly interfering with the desired properties.

The synthetic lung surfactant compositions of the invention are simple mixtures of the dipalmitoyl phosphatidyl choline component and the fatty alcohol component. Preparation and storage of the surfactant composition may best be understood by reference to the examples set forth below.

EXAMPLE I

Synthetic lung surfactant was prepared from chromatographically pure (greater than 99%), dipalmitoyl phosphatidyl choline and hexadecanol. Both materials were purchased on the commercial market where they are available from a number of chemical supply houses. Specifically, DPPC was purchased from both the Fluka Company and Sigma Chemical Company. Hexadecanol was purchased from NuChek Prep. Company. All of the purchased materials were checked for purity by chromatographic analysis.

The lung surfactant composition was prepared as follows: 314 mg. of DPPC and 33.6 mg. of hexadecanol were dissolved in 10 ml. of 1/1 chloroform/methanol (V/V, C.P.). The dissolved materials were then transferred to a 1000 ml round bottom flask. The flask was attached to a rotary vacuum evaporator and the chloroform/methanol solvent was evaporated at 37° C. leaving the synthetic surfactant lipids in a dry, thin film on the lower half of the wall of the flask. A number of clean glass beads (5 mm diameter) and 5 ml of saline were introduced in to the flask. The flask was then stoppered and the beads were then circulated by hand by swirling until all of the lipid residue had been stripped from the wall and dispersed throughout the saline solution. The dispersing procedure was carried out at 50°–52° C. by warming under running tap water. After the initial suspension of the lipids in saline an additional 18 ml of saline was added to make a total volume of 23 ml. The resultant suspension had a concentration of about 15 mg. of lung surfactant per ml. The suspension was transferred to a 30 ml syringe for dispensing.

Upon standing the suspension settled in about 5 minutes, but it could be readily redispersed by swirling even after a week of storage at 5° C. The suspension is capable of being preserved indefinitely when frozen at −70° C.

A portion of the above-noted preparation was re-suspended in distilled water to check its properties. The appearance of the suspension was like that in saline i.e. it was pure white in color, had no taste or odor and was completely bland and nonirritating to the tongue and mucus membranes of the mouth and nose.

EXAMPLE II

In an alternate method the synthetic lung surfactant may be prepared according to the following:

Synthetic 1,2 dipalmitoyl-sn-3 glycerophosphorylcholine (99% pure) (DPPC) may be obtained from Sigma Co., St. Louis, Mo. or Applied Science Labs. State College, Pa. and checked by thin layer and gas-liquid chromatography for contaminants and degradation products. It can be used only if it is at least 99% pure by chromatography. Phosphorus must be between 3.9 and 4.1% by weight. Specific rotation should be $\alpha_{20}{}^{D}+5.7°$ (10% in chloroform).

Synthetic n-hexadecan-1-ol (>99% pure) may be obtained from Nu Chek Prep Co., Elysian, Minn., and checked by gas-liquid chromatography for other fatty

alcohols. It is acceptable if it contains not more than 1% of other fatty alcohols.

DPPC and the fatty alcohol are dissolved in a ratio of 9:1 by weight in redistilled chloroform to give a solution containing 1.125 grams total in 20 ml. This solution is placed in a sterile Virtis 150 ml lyophilization flask and the chloroform completely removed by rotary vacuum evaporation so as to deposit the lipids in a film on the bottom one third of the flask. 100 ml of sterile 0.10 N sodium chloride (SUP) solution is added and the lipids suspended by intermittent sonication (Branson sonifier 185, large probe, scale setting 50) at room temperature until the suspension is uniform to inspection. Care must be taken that the temperature of the solution does not exceed 35° C. Half of this solution is then shelled (frozen) on the wall of each of two sterile 300 ml Virtis flasks, using a dry-ice alcohol bath, and subsequently lyophilized. The residue containing 802 mg. surfactant and 421 mg. sodium chloride, total weight 1,224 mg. in each flask is pulverized with a spatula and then transferred to 7 sterile 10 ml. vacuum vials, 175 mg. of pulverized product in each. The vials are evacuated and stoppered with vacuum-tight rubber seals, with a Virtis apparatus, and capped. The vials may be stored at 5° C. or below until needed.

Administration of the Lung Surfactant Compositions

The compositions, prepared as noted in Examples I and II above, are intended for administration directly into the lungs of the distressed subject.

In the case of preparation according to Example I, the frozen composition is allowed to warm to ambient temperature, at which time, the lipids are redispersed in the saline medium by swirling. The redispersed compositions and saline are then simply introduced directly into the lungs via an endotracheal tube. A dose rate of about 7.5 ml./kg. (112 mg./kg.) of subject body weight is adequate.

In the case of the preparation according to Example II, it has already been noted that the material is redispersed shortly before use. More specifically, 15 minutes before use, the material is reconstituted with 10 ml. distilled water. A kit is provided which contains:

1 vial of surfactant, 115 mg.-sodium chloride 60 mg.;
1 ampoule of sterile distilled water, 15 ml. and an ampoule knife;
1 30 ml. disposable syringe, 3-way stopcock, and two 20 gage needles;
1 alcohol gauze pad, 2×2;
A tank of medical grade oxygen is to be available.

The vacuum vial is uncapped and the rubber seal cleaned with alcohol. The ampoule is cleaned, scored, and opened and 10 ml of the distilled water aspirated into the 30 ml. syringe. The seal of the vaccum vial is punctured so that the distilled water is drawn into the surfactant. A second 20 gage needle is introduced through the seal, so that the suspension of surfactant can be passed vigorously at least 5 times between the vial and the syringe and finally into the syringe. 10 ml. of oxygen is drawn in, via the side port of the stopcock.

When the subject's weight is known, the suspension is re-mixed and all but 7 ml./kg. is expelled into the vial. The stop cock is closed. The remainder, containing 80 mg. surfactant/kg. (about 15 times the normal amount of alveolar DPPC), is shaken well in the syringe with the oxygen. Suspension, foam, and oxygen are administered via a cuffed endotracheal tube and followed by vigorous resuscitation.

Testing

The synthetic surfactant compositions are tested both in vitro and in vivo. Of course, several requirements of the synthetic surfactant compositions are inherently satisfied because of the components themselves. Specifically, since the DPPC and fatty alcohol are secured from sources which have synthesized the components and the components are tested for assured purity, no proteins are present in the compositions. Thus the chance of antibody reaction by the treated subject is eliminated. Secondly, since the components have not been derived from animal sources, there is essentially no chance for contamination by bacteria or viruses. Thirdly, since the components are secured in a highly pure state, it is easy to prepare standardized and therefore reproducible mixtures from batch to batch of surfactant. Thus, quality control is greatly simplified. Finally, since both components occur naturally, although not associated, within animal tissues, metabolic pathways for eventual elimination are already established and there is no introduction into the subject of biologically foreign substances.

As to the specific properties required of such surfactant compositions and set forth hereinbefore, a relatively simple in vitro test has been devised. This test is a "shake test" modified from a procedure devised by the present inventor for the purpose of testing for natural surfactant in amniotic fluid. This test was originally disclosed in the New England Journal of Medicine 286 pp. 1077-1081, 1972.

The test is as follows:

A sample of the carefully mixed synthetic surfactant composition prepared according to Example II containing about 400 micrograms of the surfactant is placed in a 20 ml. culture tube. 2 ml. of saline is added, and the tube is tightly capped with a screw cap. The capped tube is immersed in a water bath held at 37° C. for a time (5 minutes) sufficient to equilibrate the temperature of the sample with that of the bath. The tube is then removed and shaken vigorously by hand for 15 seconds and then replaced into the bath.

The presence of a copious foam at the meniscus confirms that the surfactant components are absorbed from the liquid phase into the surface and create a film thereon. If the bubbles are tiny and remain for 15 minutes or more, the test confirms that the surface film is stable and maintains a low surface tension.

All samples prepared according to the invention compositions, passed the "shake test". Although the test is quite simple, it has been shown to correctly assay several of the properties required by lung surfactant compositions.

The surfactant compositions may also be tested in vivo on prematurely delivered lambs at 120–130 days gestation. At this stage of gestation endogenous lung surfactant is absent and respiratory function is inadequate.

When a 90% DPPC-10% hexadecanol suspension was introduced directly into the bronchi at a dosage of approximately 90 mg/kg of body weight, subsequent arterial blood analysis indicated good CO_2 and O_2 exchange. Rapid lung expansion was also noted.

I claim:

1. A mammalian lung surfactant composition consisting essentially of dipalmitoyl phosphatidylcholine in admixture with a fatty alcohol.

2. The composition of claim 1 wherein the fatty alcohol has from about 14 to 18 carbon atoms.

3. The composition of claim 2 wherein the fatty alcohol is hexadecanol.

4. The composition of claim 2 wherein the fatty alcohol is oleic alcohol.

5. The composition of claim 1 wherein the dipalmitoyl phosphotidyl choline constitutes a major percentage by weight of the composition and wherein the fatty alcohol constitutes a minor percentage.

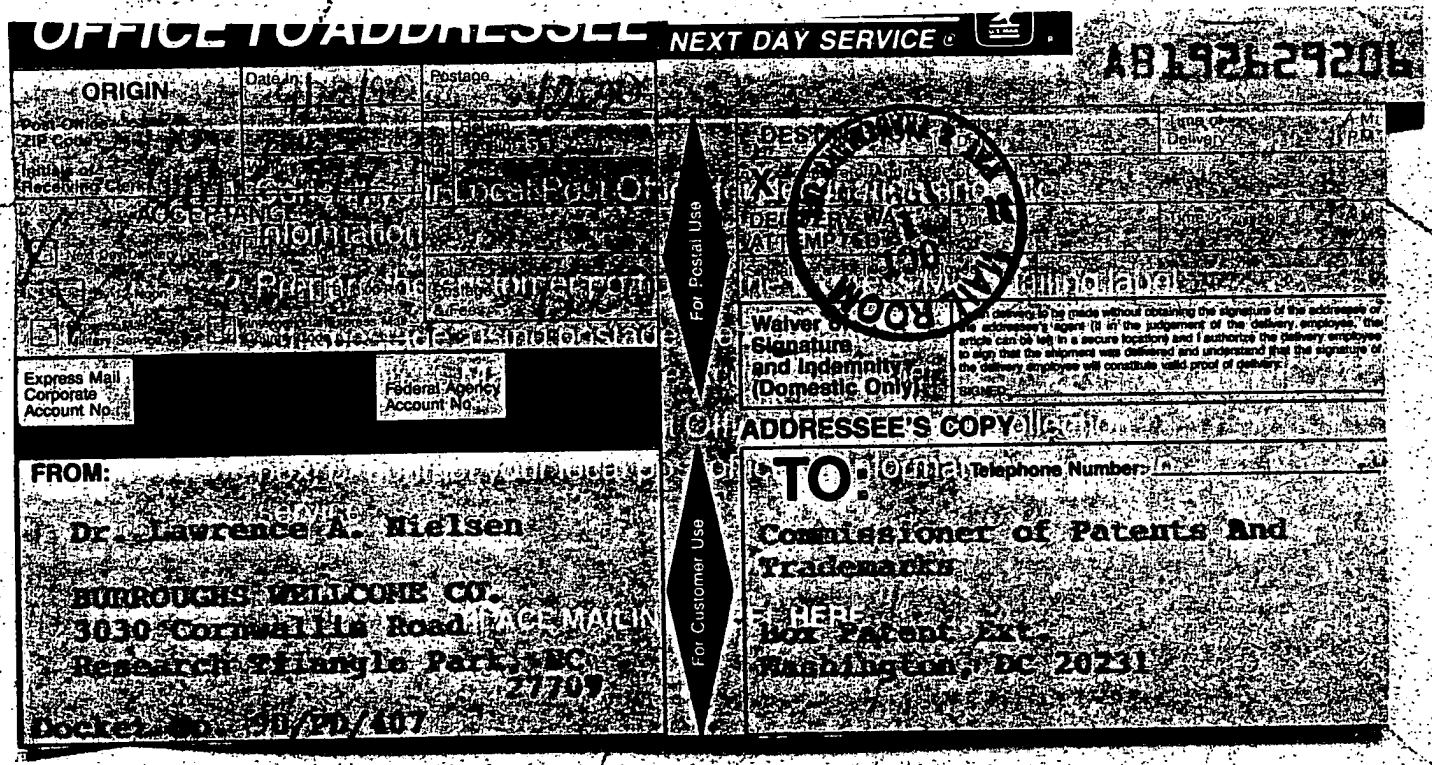
6. The composition of claim 5 wherein the fatty alcohol is present in the range of about 6 to 18% by weight and the dipalmitoyl phosphatidyl choline is present in the range of about 82 to 94% by weight.

7. A composition for administration into mammalian alveolar spaces comprising a suspension of dipalmitoyl phosphatidyl choline and hexadecanol in saline solution.

8. A method for treating respiratory distress syndrome in mammals wherein natural lung surfactant normally produced by the mammal is absent or deficient, comprising introducing into the alveolar spaces a quantity of a composition consisting essentially of a major amount of 1,2 dipalmitoyl-sn-3-glycerophosphoryl choline in admixture with a minor amount of a fatty alcohol.

9. The method of claim 8 wherein the fatty alcohol is n-hexadecan-1-ol.

* * * * *



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